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INHIBITION OF REPORTER GENES BY SMALL INTERFERING RNAS IN CELL CULTURE AND LIVING FISH

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RNA interference is a mechanism for silencing specific genes. It has been applied in cell culture to inhibit expression of genes involved in disease including viral genes as recently shown for the fish pathogenic rhabdovirus viral haemorrhagic septicaemia virus or VHSV (Bohle et al., 2011). But evidence of specific siRNA inhibition in living fish is still needed. Using the small interfering RNAs (siRNAs), messenger RNA (mRNA) can be targeted resulting in degradation of targeted transcript or translational repression. Reporter genes such as luciferase and green fluorescence protein (GFP) can be used to observe the knock down effect by siRNAs designed to target these reporters. One aim of this project is to verify the specific knock down effect of siRNAs in cell culture and in living fish and to establish easy-read out models for testing the effect especially *in vivo*. Cell culture from human embryonic kidney HEK293t cells was used because they are easy to transfect and generally show high expression of transfected genes. Various types of fish including albino trouts and transparent fish were used as animal models to get better visualization of reporter gene expression. High variability of reporter gene expression was found between individual fish but it seems that in glass catfish, siRNAs are able to reduce reporter gene expression in the muscle showing that it is possible to use siRNA as technology to target genes locally in living fish. In parallel experiments, which will not be reported here, we examine the delivery of siRNAs using pharmacological formulations in order to achieve systemic delivery and knock down effect.